Association of glomerular hyperfiltration with serum chemokine levels and metabolic features in prepubertal children with overweight/obesity

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ABSTRACT

Background and aims: Glomerular hyperfiltration (GH) is proposed as one of the earliest events in obesity (OB)-associated renal disease. Children with GH and type-1 diabetes showed increased chemokine levels. Chemokine associations with glomerular filtration rate (GFR) and metabolic features in prepubertal children with overweight (OW)/OB are unknown. Methods and results: Cross-sectional study. 75 prepubertal children (aged: 9.0 ± 1.7 years) with OW/OB were studied. Clinical and metabolic characteristics (including non-esterified fatty acids, NEFA) and GFR (combined Zappitelli equation) were assessed. GH was defined as GFR >135 ml/min.1.73 m2. Serum levels of regulated on activation, normal T cell expressed and secreted (RANTES)/CCL5, interleukin-8 (IL-8)/CXCL8 and monokine-induced by interferon-g (MIG)/CXCL9 were measured by ELISA. Age- and sex-adjusted correlations and differences were tested. 48% of the cohort was female and 13% were OW, 54% OB and 33% severe OB. Prepubertal children with GH showed lower z-BMI (-12%), NEFA (-26%) and uric acid (-22%) than those without GH (all $p < 0.05$). Similarly to high sensitivity C-reactive protein (hsCRP), there were no differences in serum chemokines between children with GH or not (all $p > 0.05$). Adjusted correlations were significant for RANTES and z-BMI (r Z 0.26; $p < 0.05$) and for MIG with z-BMI (r Z -0.26; $p < 0.05$) and with NEFA (r Z 0.27; $p < 0.05$). *Conclusion:* GH was not associated with higher chemokine levels in prepubertal children with OW/OB. Decreased rather than elevated GFR values were correlated with obesity and worse metabolic profiles. Chemokines levels in children with severe OB suggest a regulation of the immune response. Follow-up studies are needed to address the clinical implications of these findings.

Introduction

Childhood obesity (OB) is a growing health problem worldwide [1]. Children with overweight (OW) and/or OB are at increased risk for developing several comorbidities during adult age such as type 2 diabetes and renal and cardiovascular diseases, among others [2,3]. In the development of obesity-associated renal disease an early stage of elevated glomerular filtration rate (GFR) or glomerular hyperfiltration (GH) is proposed [4]. Whether elevated GFR is a relevant clinical sign indicating early impairment in renal function in children with OB is a matter of debate [5e9]. Some reports described

increased GFR in children with OB in comparison with normal-weight children; however, others showed decreased GFR levels [5e9]. In part, controversy arises from the difficulties associated with the definition of GH. Some authors used an estimated GFR calculated by the modified Schwartz equation, which uses only serum creatinine concentration, while others used equations with cystatin C concentration alone or with a combination of both creatinine and cystatin C levels [5,7,8]. Also, different authors employed different cut off points of GFR, although a value >135 ml/min.1.73 m2 is frequently used in the literature [5,10]. In adults with OB and obesity related glomerulopathy (ORG), GH is commonly associated with metabolic alterations and increased inflammatory markers [4]. Whether such associations are also found in prepubertal children with OB have not been fully described. Lee et al. [11] showed increased triglycerides and markers of insulin resistance in adolescents with GH. In addition, Di Bonito et al. [12] showed that OB children (6e16 years) with an altered GFR (defined as >80th percentile or <20th percentile) were at increased risk of presenting elevated blood pressure (BP), impaired fasting glucose (IFG) and elevated white blood cell counts. Non-esterified fatty acids (NEFA) are at the hallmark of metabolic abnormalities and endothelial dysfunction in adults with OB and insulin resistance [13]. Impaired lipolysis and elevated NEFA in adults with OB were linked to ectopic fat deposition in several organs, including the kidneys [14]. “Fatty” kidney could be one possible explanation to the impairment of renal function in OB [14]. Indeed, a recent study showed that single nucleotide polymorphisms (SNPs) predisposing to ectopic fat accumulation were associated with lower GFR values in OB children with non-alcoholic fatty liver disease [15]. NEFA levels and its association with GFR and inflammatory markers in prepubertal children has been scarcely studied. A link between inflammatory markers and GH was suggested by a positive correlation between GFR and myeloperoxidase activity in a Portuguese cohort of prepubertal children with OW/OB [16]. However, most of the studies regarding GH and chemokines were carried out in children and adolescents with type 1 diabetes. In particular, children with type 1 diabetes and GH showed elevated urinary excretion of monocyte chemoattractant protein (MCP)-1, regulated on activation, normal T cell expressed and secreted (RANTES) and eotaxin in comparison with those without GH [17]. T lymphocyte chemokines, like RANTES/CCL5 and interleukin-8 (IL-8)/ CXCL8, are secreted by adipose tissue and are proposed as important mediators

in the cross-talk between OB and cardiovascular disease [18e20]. Another chemokine relevant to vascular health is the monokine induced by interferon- g (MIG)/CXCL9 which is postulated as an important mediator in hypertension and cardiovascular disease [21]. In children with OB, levels of RANTES, IL-8 and MIG decreased with body weight reductions or BP changes during intervention studies [22,23]. Then, these chemokines may be involved in early changes of the GFR in prepubertal children with OB. The aim of the present study was to characterize the correlations of serum chemokines with GFR and metabolic alterations in a cohort of prepubertal children with OW/ OB. A secondary analysis was performed to explore differences between children with or without severe OB.

Methods

Sample size, study design and population

Sample size was calculated in the base of a previous study on correlations between chemokines and GFR in children and adolescents [17]. As it was done in pediatric patients with type 1 diabetes, a-priori we multiplied the result by 1.5 for final sample size estimation. The number of patients to be studied with an effect size of 0.381, 80% power and two-tailed significance level of 0.05, multiplied by 1.5 was 73. The studied cohortwas part from an ongoing prospective study which consecutively recruited every child between 5 and 17 years with OWor OB who consulted to the Pediatric Diabetes and Nutrition Service of the Complejo Médico Churruca Visca, Buenos Aires, Argentina, during April 2017 to October 2018. Out of the 183 patients recruited, we filtered the prepubertal ones (nZ109). Fromthese, a simple random sample of 80 patients was used for the analysis. At the time of measuring high sensitivity C reactive protein (hsCRP), 5 patients had to be excluded in the basis of a value >10 mg/l. Thefinal numberof prepubertal children included in this cross-sectional study was 75. All patients, including the group of normal weight children, and their parents or responsible adult signed the assent and consent forms, respectively, before participation in the study. The protocol was approved by the Ethics Committee of the Complejo Médico Churruca-Visca. The general exclusion criteria for the study were diabetes, hypothyroidism or treatment with levothyroxine, concomitant

autoimmune or liver pathologies, an urinary albumin-creatinine ratio >30 mg/g, clinical signs of an infectious disease or a hsCRP >10 mg/l.

Clinical and laboratory measurements

All the patients underwent a clinical interview in the presence of their parents or responsible adult in which height and weight were determined without shoes and wearing light clothes. Body mass index (BMI) was calculated and BMI classification was performed according to the z-score from the World Health Organization (WHO) [24]. Severe OB was defined by a z-BMI > þ3.0. BP was measured twice after a 5 min rest in the sitting position and the average of both measurements was recorded. The American Academy of Pediatrics (AAP) 2017 guidelines were used to calculate BP z-scores and to define elevated BP [25]. Waist circumference (WC) was measured at the mid-point between the iliac crest and the 10th rib and elevated WC classified as >90th percentile of the Bogalusa Heart Study [26]. After a 12 h overnight fast, blood samples were drawn and general lab measurements were carried out within a 24 h period. Serum samples were frozen at └ 70 ┘ C for the hsCRP and chemokines measurements. General laboratory testswere carried out in aUnicel DxC 800 (Beckman Coulter, USA) autoanalyzer by standardized methods. Cystatin C concentrationwas measured with an immunoturbidimetric assay (Diazyme, USA) and NEFA by a colorimetric assay (Randox, UK) according to manufacturer indications. HsCRP was measured by a nephelometric assay (Immage, Beckman Coulter, USA). GFR was estimated by the combined creatinine and cystatin C Zappitelli formula andGHwas defined in presence of a GFR >135 ml/min.1.73 m2 [27]. In order to verify the GH cut-off suggested by bibliography [28,29], 26 samples of prepubertal normal weight children were tested for creatinine and cystatin C and only one patient showed an GFR outside the defined reference range (90e135 ml/ min.1.73 m2). These 26 children (44% female, aged: 9 2 years) were attended in the same Hospital service as the patients and, besides being normal-weight [z-BMI: median (Q1dQ3), 0.07 (-0.67 d0.62)] met the same exclusion criteria as the patients with OW/OB. The range of GFR from these children was between 98 and 136 ml/min.1.73 m2, with a median at 117 ml/min.1.73 m2 and Q1dQ3 Z 111e126 ml/min.1.73 m2. Serum levels of RANTES/CCL5, MIG/CXCL9 and IL-8/ CXCL8 were measured by ELISA (R&D systems, USA). Cardiometabolic

risk factors were categorized according to the definition of metabolic syndrome of Cook et al. [30] modified by the Sociedad Argentina de Pediatría. The following cut-offs were used: abdominal OB, WC > 90th percentile; elevated BP, systolic or diastolic BP > 90th percentile; impaired fasting glucose (IFG), glucose > 100 mg/dl; hypertriglyceridemia (HTG), TG > 110 mg/dl and low HDL-C, HDL-C < 40 mg/dl.

Statistical analyses

Variables were tested for normality by the ShapiroeWilks test. Normally distributed variables are shown as mean ± standard deviation (sd) and skewed distributed variables as median (Q1eQ3). According to data distribution, Pearson or Spearman correlation tests were used. The different groups were compared by Student T test or ManneWhitney U test for normally or skewed distributed variables, respectively. Following log-transformation of skewed variables partial correlation test or ANCOVA was used to adjust by age and sex. Residuals of multivariate tests were checked for normality to evaluate the models. Categorical variables were compared with the Fisher Exact test. SPSS 25.0 (IBM, USA) was used for statistical analyses. Tests were considered statistically significant with a significance value (p) < 0.05.

Results

Population characteristics and evaluation of renal function

The studied population comprised 75 prepubertal children (48% female) of which 13% showed OW, 54% OB and 33% severe OB. The overall prevalence of metabolic syndrome components were: abdominal OB: 73%, low HDL-C: 57%, HTG: 39%, elevated BP: 23%, and IFG: 9%. Metabolic syndrome was diagnosed in 32% of the children and GH was found in 16%. Clinical and laboratory characteristics according to the presence of GH are shown in Table 1. Prepubertal children with GH showed lower z-BMI, NEFA and uric acid than those with normal GFR. GFR correlated with age (r Z -0.32; p Z 0.005), NEFA (r Z -0.30; p Z 0.011) and uric acid levels (r Z -0.29; p Z 0.011). These correlation were independent of sex and age and sex, accordingly. Besides its inverse correlation with GFR, uric acid levels correlated with the number of cardiometabolic risk factors (r Z 0.37; p < 0.001),

LDL-C (r Z 0.30; p Z 0.010), TG/ HDL-C (r Z 0.28; p Z 0.015), NEFA (r Z 0.39; $p < 0.001$) and hsCRP (r Z 0.29; p Z 0.014). These correlations, except that with TG/HDL-C, remained statistically significant when adjusting by age and sex.

Serum chemokine levels and its correlation with renal function and metabolic characteristics

Similarly to the inflammatory marker hsCRP (Table 1), there were no differences in serum chemokines between children with GH or not [RANTES: 27.2 (22.4e29.0) vs. 26.9 (23.6e30.0) ng/ml; MIG: 420 (329e523) vs. 454 (276e686) pg/ml; and IL-8: 551 (311e782) vs. 630 (406e922) pg/ml, respectively, all $p > 0.05$]. MIG and IL-8 were both inversely correlated with GFR (r Z -0.27, p Z 0.035; and r Z -0.25, p Z 0.022, respectively). These correlations were no longer significant when tests were adjusted by age and sex ($p > 0.05$). RANTES levels were not correlated with GFR ($p > 0.05$). Correlations of serum chemokine levels with z-BMI and NEFA are shown in Fig. 1. Strikingly, while RANTES was directly correlated with z-BMI, MIG was inversely correlated with z-BMI and positively with NEFA (Fig. 1). IL-8 levels were closely correlated with MIG levels (r Z 0.78; $p < 0.001$), and IL-8 correlations with z-BMI and NEFA mirrored those of MIG (Fig. 1). In analyses adjusted by age and sex, only RANTES correlation with z-BMI and both MIG correlations remained statistically significant (all $p < 0.05$). None of the other metabolic characteristics were correlated with the serum chemokine levels. HsCRP levels were significantly correlated with z-BMI (r Z 0.45; $p < 0.001$) and this correlation remained significant even when adjusting by age and sex ($p < 0.05$).

Subgroup analysis according to severe obesity

Prepubertal children with severe OB were younger and showed a worse cardiometabolic profile (Table 2). Strikingly, children with severe OB did not present GH which was exclusively seen in children without severe OB (Table 1). Children with severe OB presented higher levels of hsCRP but lower levels of the chemokines IL-8 and MIG. Differences in hsCRP and MIG remained statistically significant in models adjusted by age and sex (Table 2). Adjusted differences were also significant for HOMA-IR and TG.

Discussion

The present study shows that serum chemokines concentrations were not different between prepubertal children with or without GH. Moreover, children with GH were less obese and did not showed higher levels of cardiometabolic risk factors. In fact, children with severe OB did not show a higher GFR, in spite of being younger. Altogether, these results suggest the existence of a pool of OB children with an early decline of GFR in which GH could have happened at even younger ages, as declining GFR values in childhood OB were also noticed by others [9,15]. As an alternative, the present definition of GH (GFR > 135 ml/min.1.73 m2) might be limited to detect early renal impairment. Whatever the case, this insight is clinically relevant as prepubertal children with OW/OB developing early alterations in renal function would not be identified by current biochemical markers. In this cohort of prepubertal children there were no correlations between GFR and z-BMI. Moreover, instead of positive, negative correlations of GFR with NEFA and uric acid were seen. Both elevated NEFA and uric acid are not only markers but also proposed mediators of cardiometabolic derangements in OB. Uric acid was positively correlated with cardiometabolic risk factors, the TG/HDL-C ratio, LDL-C, NEFA and hsCRP, similarly to what it was reported by other studies in prepubertal children with OB [31]. Thus, our data supports the notion that OB and its metabolic alterations could lead to a reduction of GFR rather than to an increase in prepubertal children. This result contrasts with the studies that used different GFR equations, like Bouvet or modified Schwartz [5,9,27]. In particular, Bouvet equation includes weight as a variable and then higher but not lower GFR values in OB children could be expected [5]. On the other hand, the use of Schwartz equation could lead to bias related to body distribution of fat and lean mass and its influence on creatinine levels [9]. Although other sources of variation in serum cystatin C levels, like increased secretion by adipose tissue cannot be dismissed [32], our results support those of a Portuguese cohort of 313 prepubertal children (48% with OW/OB) [7]. In the latter, not only GFR using Zappitelli equation was negatively correlated with z-BMI, but also 24h creatinine clearance [7]. Nonetheless, the impact of OB over GFR in childhood remains to be determined in longitudinal studies. Prepubertal children with severe OB showed a worse cardiometabolic profile and, paradoxically, elevated hsCRP and decreased MIG levels. In this point, it should be noticed that while hsCRP were positively correlated with z-BMI, MIG did in an opposite way. Then, the IL-1b d IL-6 d CRP axis could reflect the

developing inflammatory state set up by the onset and progression of fat accumulation, as it was also noticed in cross-sectional and intervention studies involving prepubertal children [33e36]. On the other hand, the inverse correlation between z-BMI and MIG could be related to an adaptative counterregulation involving the adipose tissue regulatory T cells (Treg) [37]. This adaptative Treg response was increased in OB vs. lean humans and was more relevant in subcutaneous than visceral adipose tissue [38,39]. Such specific responses in fat depots is relevant to the interpretation of data on prepubertal children with OB. Before puberty, gain of subcutaneous fat mass is more relevant, while from puberty onset and onward visceral fat accumulation is more significant [40,41]. Indeed, when comparing prepubertal children with or without OB differences in subcutaneous adipose tissue were quantitatively more significant than those in visceral adipose tissue [36,42]. Also, subcutaneous rather than visceral adipose tissue was correlated with reduced insulin sensitivity and ectopic fat accumulation before puberty [36,42,43]. On the other hand, MIG negative correlation with NEFA could be at some point attributed to the known effects of IFN-g on lipolysis [44]. In regards to IL-8, its correlations with metabolic characteristics resembled those of MIG and an inverse correlation between IL-8 and z-BMI was also reported in a cohort of prepubertal children [42]. Although IL-8 expression was demonstrated in adipose tissue, the correlation between MIG andIL-8 was independent of z-BMI in our study (data not shown). Thus, another source of MIG and IL-8 (like monocytes) could be suspected in prepubertal children with OB. Indeed, IFN-g promoted the secretion of IL-8 and MIG by monocytes in synergy with tumor necrosis factor (TNF)-a [45]. The positive correlation between RANTES and z-BMI is in line with T cell infiltration of adipose tissue as a consequence of fat mass expansion [38]. T cell infiltration is proposed as one of the earliest events in OB [46]. Thus, elevation of RANTES could be related to an adaptative response to OB in children, as RANTES exhibited antiapoptotic activities in adipose tissue macrophages [47]. Nonetheless, elevated RANTES levels were correlated with arterial stiffness, an early marker of endothelial dysfunction, in healthy prepubertal children [48]. Then, RANTES levels could mirror maladaptative responses in OB and it might be related with its cardiometabolic consequences. The cross-sectional design of our study did not allow us to draw conclusions on mechanisms and our hypothesis to explain the correlations observed are speculative. Nonetheless, few studies focused in

the relationship between chemokines, NEFA and GFR in prepubertal children with OB. Moreover, children with severe OB represent a group of understudied individuals, especially before pubertal development. In the present cohort, severe OB was present in 33% of the children with z-BMI values ranging: 3.03e6.72. The evaluation of these patients by separate gave further support to the notion of declining GFR (and increasing uric acid levels) in children with OB as possible early events in the development of renal disease. Future studies will address if control of uric acid levels may help preserve renal function in childhood OB. On the other hand, the relatively low number of patients included, could be considered a shortcoming and raises the possibility to have missed relevant results. Among the strengths of the study there are two to be highlighted: a) the exclusion of patients with clinical or biochemical signs (hsCRP > 10 mg/l) of infectious or inflammatory conditions that minimized the possibility of bias in chemokine levels due to concomitant pathological processes and b) the use of sex and age-adjusted analysis to draw the main conclusions of the study. In conclusion, in prepubertal children with OW/OB serum levels of RANTES, IL-8 and MIG were not correlated with the GFR. Increasing z-BMI and worse metabolic profiles were associated with decreased rather than elevated GFR values. While RANTES was positively correlated with z-BMI, MIG showed an inverse correlation. Follow-up studies are needed to address the clinical implications of these findings.

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412

413 **FIGURES**

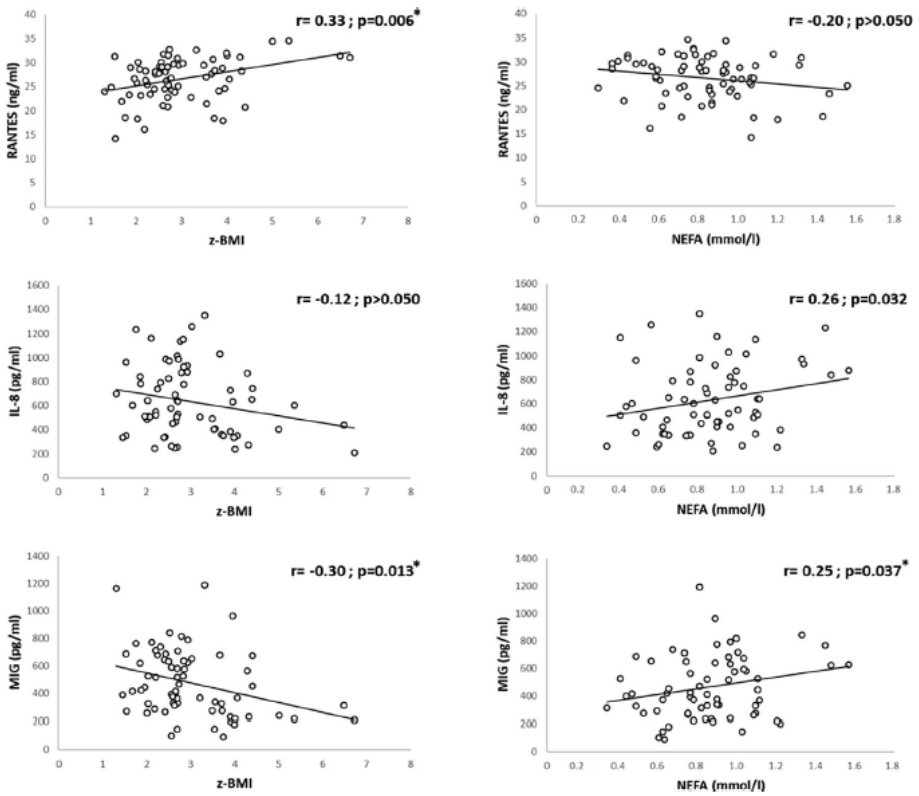

Figure 1 Correlations between serum chemokines and z-BMI and NEFA (n = 75). NEFA, non-esterified fatty acids; RANTES, regulated on activation, normal T cell expressed and secreted; IL, interleukin; MIG, monokine induced by interferon γ. Coefficients and significance levels shown are those of univariate tests. *Remained significant in tests adjusted by age and sex.

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TABLES

Table 1 Characteristics of prepubertal children with OW/OB according to the presence of GH (n = 75).

	GH (n = 12)	Normal GFR (n = 63)	p
Age (years)	8.2 ± 1.2	9.2 ± 1.7	0.214
Female sex (n,%)	8 (66)	28 (44)	0.212
z-BMI	2.5 (2.0–2.6)	2.8 (2.4–3.7)	0.016[a]
Severe OB (n,%)	0 (0)	25 (40)	0.005
z-SBP	0.51 (-0.19 — 0.87)	0.62 (-0.19 — 1.08)	0.470
z-DBP	0.32 (-0.11 — 0.57)	0.47 (-0.31 — 0.93)	0.509
Elevated BP (n,%)	2 (17)	16 (25)	0.719
WC (cm)	75 ± 7	84 ± 13	0.054
Glucose (mmol/l)	5.0 ± 0.4 [90 ± 7]	5.2 ± 0.4 [93 ± 7]	0.326
HOMA-IR	2.7 (1.4–4.7)	2.2 (1.6–3.8)	0.740
TG (mmol/l)	1.1 (0.6–1.2) [97 (57–104)]	1.2 (0.8–1.6) [103 (69–140)]	0.308
TC (mmol/l)	4.5 ± 0.6 [159 ± 22]	4.7 ± 0.8 [166 ± 29]	0.403
HDL-C (mmol/l)	1.03 (0.94–1.23) [36 (33–43)]	1.09 (0.9–1.4) [38 (33–48)]	0.115
LDL-C (mmol/l)	2.9 ± 0.5 [103 ± 17]	2.9 ± 0.7 [103 ± 26]	0.996
Non-HDL-C (mmol/l)	3.5 ± 0.7 [121 ± 23]	3.6 ± 0.8 [125 ± 28]	0.641
MetS (n,%)	3 (25)	21 (33)	0.741
NEFA (mmol/l)	0.69 ± 0.27	0.87 ± 0.26	0.027[a]
Uric acid (μmol/l)	220 ± 42 [3.7 ± 0.7]	267 ± 59 [4.5 ± 1.0]	0.013[a]
Creatinine (μmol/l)	34 (32–38) [0.38 (0.36–0.43)]	42 (38–47) [0.47 (0.43–0.53)]	0.002[a]
Cystatin C (mg/l)	0.65 ± 0.08	0.83 ± 0.08	<0.001[a]
GFR (ml/min.1.73 m^2)	144 ± 10	113 ± 12	<0.001[a]
hsCRP (mg/l)	1.5 (0.4–2.9)	2.1 (1.1–5.5)	0.165

OW, overweight; OB, obese; BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; BP, blood pressure; WC, waist circumference; TG, triglycerides; TC, total cholesterol; HDL, high density lipoprotein; LDL, low density lipoprotein; MetS, metabolic syndrome; NEFA, non-esterified fatty acids; GFR, glomerular filtration rate (calculated with combined Zappitelli equation).

[a] Differences remained statistically significant in models adjusted by age and sex. Traditional units are in brackets. Variables are expressed as mean ± standard deviation or median (Q1–Q3), according to data distribution.

Table 2 Comparison of patient's characteristics with or without severe obesity.

	Severe OB (n = 25)	OW/OB (n = 50)	p
Age (years)	8.4 ± 1.6	9.4 ± 1.6	0.014
Female sex (n,%)	8 (32)	28 (56)	0.056
z-BMI	3.9 (3.6–4.4)	2.5 (2.0–2.7)	<0.001[a]
z-SBP	0.73 (0.15–1.39)	0.55 (-0.16 — 1.03)	0.057
z-DBP	0.44 (-0.22—1.40)	0.28 (-0.31 — 0.63)	0.027
Elevated BP (n,%)	10 (40)	8 (16)	0.042
WC (cm)	86 ± 15	81 ± 11	0.215
Glucose (mmol/l)	5.0 ± 0.4 [90 ± 8]	5.1 ± 0.4 [91 ± 7]	0.740
HOMA-IR	3.4 (1.8–6.7)	2.1 (1.4–3.7)	0.013[a]
TG (mmol/l)	1.6 (0.9–1.7) [139 (80–154)]	1.1 (0.8–1.3) [93 (68–112)]	0.010[a]
HDL-C (mmol/l)	1.14 (0.94–1.49) [40 (33–52)]	1.06 (0.94–1.26) [37 (33–44)]	0.356
NEFA (mmol/l)	0.78 ± 0.19	0.87 ± 0.30	0.119
MetS (n,%)	13 (52)	11 (22)	0.017
Uric acid (μmol/l)	267 ± 65 [4.5 ± 1.1]	256 ± 59 [4.3 ± 1.0]	0.416
Creatinine (μmol/l)	40 (35–42) [0.45 (0.40–0.48)]	42 (36–47) [0.47 (0.41–0.53)]	0.235
Cystatin C (mg/l)	0.80 ± 0.07	0.80 ± 0.12	0.941
GFR (ml/min.1.73 m^2)	119 ± 9	118 ± 19	0.758
hsCRP (mg/l)	4.2 (1.5–6.3)	1.5 (0.5–3.8)	0.002[a]
RANTES (ng/ml)	28.6 (24.2–31.4)	26.3 (23.7–29.1)	0.081
IL-8 (pg/ml)	438 (352–728)	640 (488–932)	0.045
MIG (pg/ml)	294 (220–536)	526 (373–693)	0.002[a]

OW, overweight; OB, obese; BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; BP, blood pressure; WC, waist circumference; TG, triglycerides; HDL, high density lipoprotein; NEFA, non-esterified fatty acids; MetS, metabolic syndrome; GFR, glomerular filtration rate (calculated with combined Zappitelli equation); RANTES, regulated on activation, normal T cell expressed and secreted; IL, interleukin; MIG, monokine induced by interferon γ.

[a] Differences remained statistically significant in models adjusted by age and sex. Traditional units are in brackets. Variables are expressed as mean ± standard deviation or median (Q1–Q3), according to data distribution.